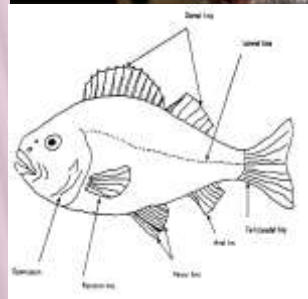
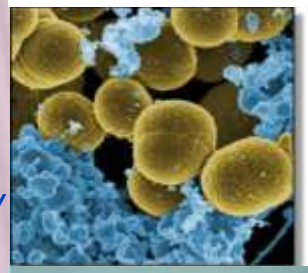
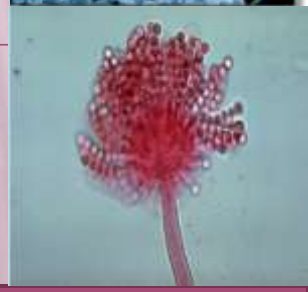
FISH AND FISHERY PRODUCTS MICROBIOLOGY– 3 (2 – 1)







MICROORGANISM IN FISH : Bacteria causing fish spoilage

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FACTORS CAUSING FOOD SPOILAGE

- Insect damage
- Physical injury
- The activity of indigenous enzyme
- Chemical changes
- The activity of MO

COMPOSITION OF CONTAMINATING MICROFLORA IN FOODS

- Gram (-) rods & coccobacilli
  - *Acinetobacter, Aeromonas, Alcaligenes, Citrobacter, Enterobacter, Escherichia, Flavobacterium, Moraxella, Proteus, Pseudomonas, Salmonella, Sewanella, & Yersinia*
- Gram (+) rods
  - *Bacillus, Brochothrix, Clostridium, Corynebacterium, Lactobacillus, & Listeria*
- Gram (+) cocci
  - *Enterococcus, Lactococcus, Micrococcus, Pediococcus, & Staphylococcus*

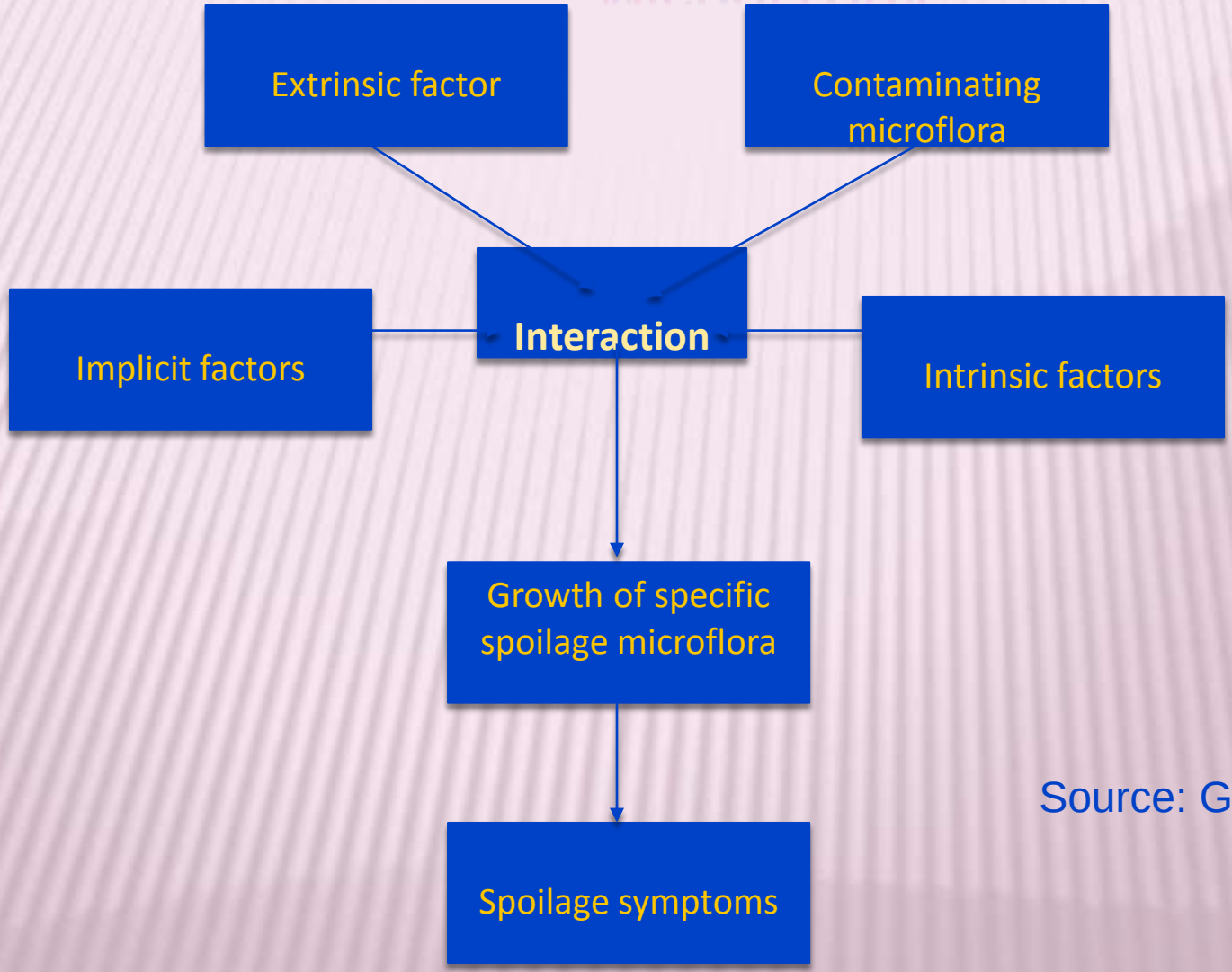
THE NATURE OF FOOD SPOILAGE

- Major reasons 4 food being rejected:
  - + Organoleptic changes → growth MO
  - + Chemical changes in food
  - + Physical damage
  - + Freezer burn
  - + 'staling' → changes Aw
  - + Ripening
  - + Presence of foreign materials
  - + Contamination with chemical agent

CHANGES IN FOODS CAUSED BY SPOILAGE MO

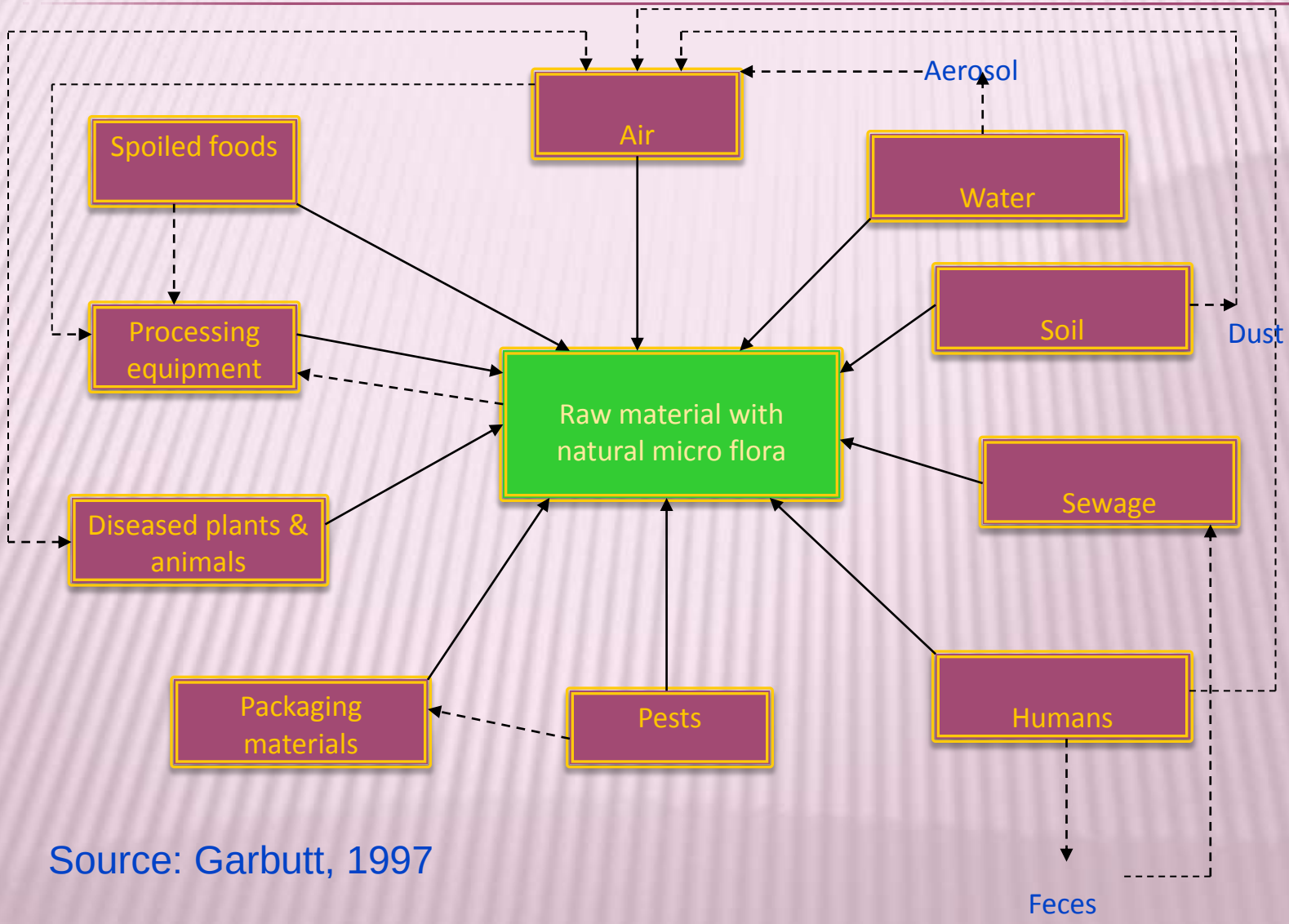
- General appearance → moldy (F) & slimy (B)
- Color → F (red/black) & B (colored); chemical changes --> greening of meat (H_2S)
- Texture → *Pseudomonas fluorescens* at fish --> prod proteinase caused tissues to soften
- Odor / flavor → MO prod chem. associated with metab. act.
- A mixture of the above

INTERACTIONS INVOLVED IN THE SELECTION OF SPOILAGE MICROFLORA



Source: Garbutt, 1997

SOURCES OF CONTAMINATION OF FOOD



Source: Garbutt, 1997

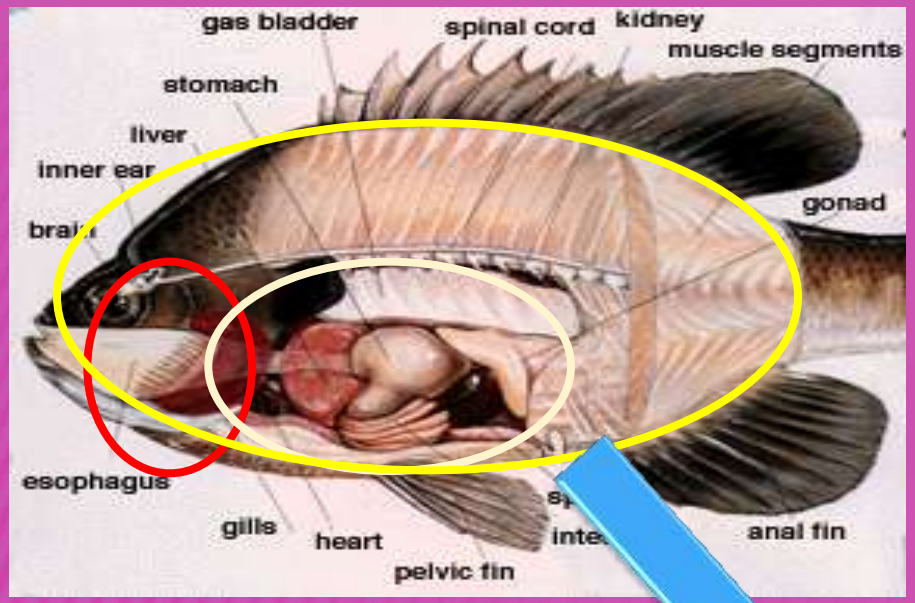
MICROORGANISMS CAUSING MOST FOOD CONTAMINATION

- Bacteria
- Parasitic worms
- Fungi
- Viruses

SPOILAGE ON FISHERY AND FISHERY PRODUCTS

IMPORTANT FACTORS CAUSING FISH SPOILAGE

- Specific as well as non-specific contamination of the live fish from environment and of products during processing.
- Growth conditions for MO due to specific intrinsic and extrinsic factors.



Microflora of temperate water fish : psychrotrophic Gram-negative, rod-shaped bacteria: *Pseudomonos, Moraxella, Acinetobacter, Shewanella, Fluvobucterium, Vibrionuceae* and *Aeroemonaduceae.*

Gram-positive : *Bacillus, Micrococcus, Clostridium, Luctobacillus* and *Corynebacterium (*varying proportions).

A **B**

What are the differences between A and B?
How is spoilage happen in fish and fish products?
Why is spoilage happen in fish and fish products?

SPOILAGE ACTIVITY OF BACTERIA

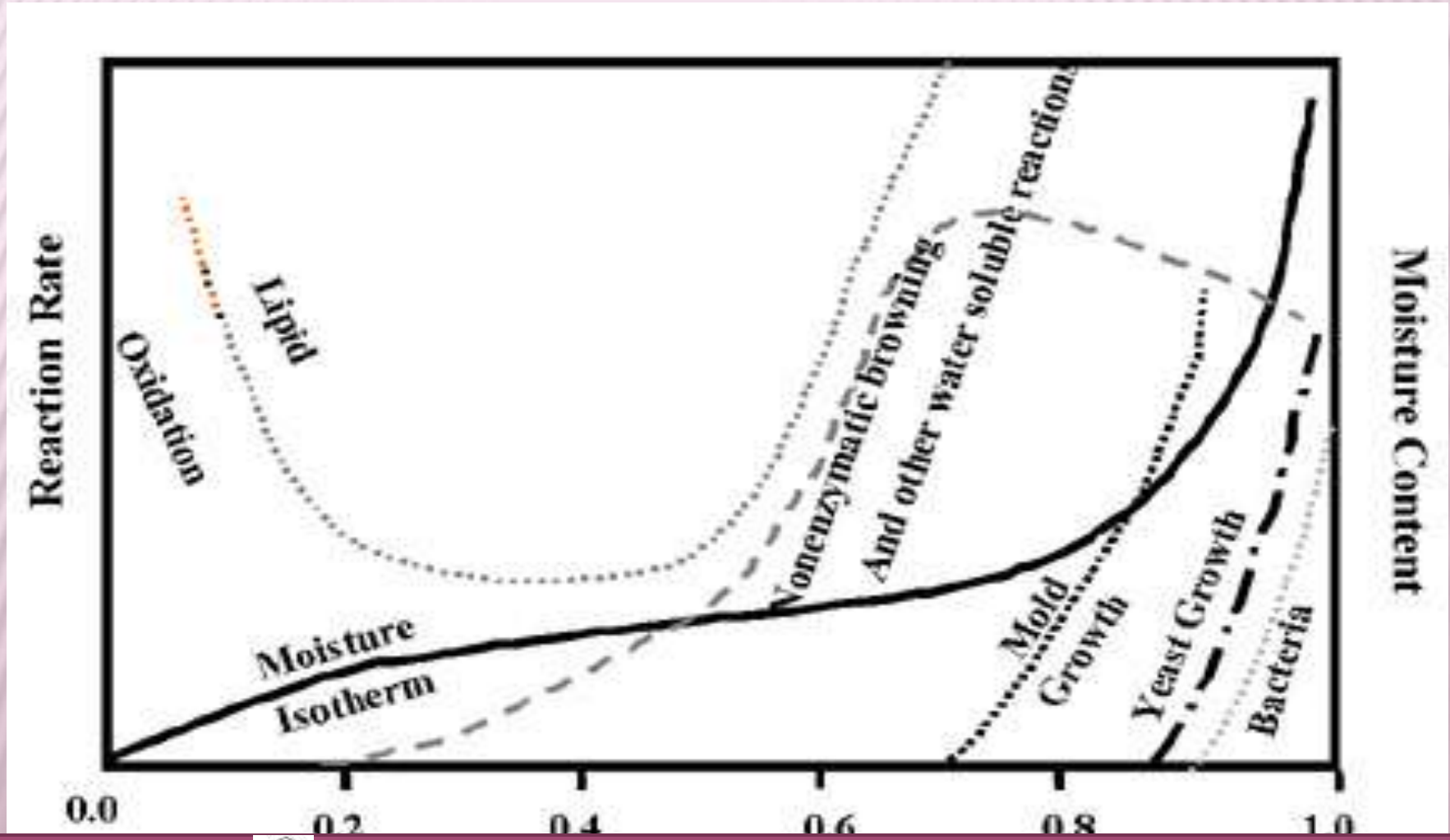
Specific spoilage bacteria	Spoilage compounds
High	*Pseudomonas (Alteronomas) putrifaciens*, *Pseudomonas (altreomonas) fluorescens* *Fluorescent pseudomonads*
Moderate	*Moraxella, Acinetobacter and Alcaligenes*
Low (Specific conditions)	*Aerobacter, Lactobacillus, Flavobacterium, Micrococcus, Bacillus and Staphylococcus*

Source: Hui, 1992 in Ghaly *et al.*, 2010

INTRINSIC FACTORS IN FISH CAUSING SPOILAGE

- The poikilotherm nature of the fish and its aquatic environment.
- A high post mortem pH in the flesh fish (usually > 6).
- The presence large amount of NPN
- The presence of TMAO
- Nutrient content
- Eh
- Aw
- Mechanical barrier

AW ON FOODS


Reaction Rate
Moisture Content
Oxidation
Lipid
Nonenzymatic browning
And other water soluble reactions
Moisture
Isotherm
Mold
Growth
Yeast Growth
Bacteria
0.0
0.2
0.4
0.6
0.8
1.0

AW RANGE OF MO

Organism	Minimum Aw	Organism	Minimum Aw
Most spoilage bacteria	0.90 – 0.91	*Staphyloccus albus*	0.88 – 0.92
Acinetobacter	0.95 – 0.98	*S. aureus*	0.83 – 0.92
Aeromonas	0.95 – 0.98	*Streptococcus*	0.92 – 0.98
Alcaligenes	0.95 – 0.98	*Vibrio parahaemolyticus*	0.94 – 0.98
Arthrobacter	0.95 – 0.98	*Halohilic bacteria*	0.75
Bacillus	0.90 – 0.99	Most yeast	0.87 – 0.94
B. cereus	0.92 – 0.95	Osmophilic yeast	0.60 – 0.78
Citrobacter	0.95 – 0.98	Most molds	0.70 – 0.80
Clostridium botulinum	0.90 – 0.98	*Xerophilic molds*	0.60 – 0.70
Type A	0.93 – 0.95	*Aspergillus*	0.68 – 0.88
Type B	0.93 – 0.96	*A. glaucus*	0.70 – 0.75
Type E	0.94 – 0.97	*A. flavus*	0.78 – 0.90
C. perfringens	0.93 – 0.97	*A. halophilicus*	0.68
Corynebacterium	0.95 – 0.98	*A. niger*	0.80 – 0.84
Enterobacter	0.95 – 0.98	*Botrytis cinerea*	0.93
Escherichia coli	0.94 – 0.97	*Debaryomyces*	0.87 – 0.91
Flavobacterium	0.95 – 0.98	*Fusarium*	0.80 – 0.92
Klebsiella	0.95 – 0.98	*Hansenula*	0.89 – 0.90
Lactobacillus	0.90 – 0.96	*Mucor*	0.80 – 0.93
Leuconostoc	0.96 – 0.98	*Penicillium*	0.78 – 0.90
Micrococcus	0.90 – 0.95	*Rhodotorula*	0.89 – 0.92
M. rosens	0.90 – 0.93	*Saccharomyces cerevisiae*	0.90 – 0.94
Pseudomonas aeroginosa	0.96 – 0.98	*S. rouxii*	0.62 – 0.81
P. fluorescens	0.94 – 0.97	*Xeromces bisporus*	0.60 – 0.61
Salmonella	0.93 – 0.96		

Sorce: Banwart, 1989

APPROXIMATE PH RANGES FOR MICROBIAL GROWTH

Organism	pH			Organism	pH		
	Min.	Opt.	Max.		Min.	Opt.	Max.
Bacteria (most)	4.5	6.5 – 7.5	9.0	Salmonella (most)	4.5 – 5.0	6.0 – 7.0	8.0 – 9.6
Acetobacter	4.0	5.4 – 6.3	-	S. typhi	4.0 – 4.5	6.5 – 7.0	8.0 – 9.0
Bacillus subtilis	4.2 – 4.5	6.8 – 7.2	9.4 – 10	S. choleraesuis	5.0	7.0 – 7.6	8.2
Clostriium botulinum	4.8 – 5.0	6.0 – 8.0	8.5 – 8.8	Serratia Marcescens	4.6	6.0 – 7.0	8.0
C. pefringens	5.0 – 5.5	6.0 – 7.6	8.5	Staphylococcus aureus	4.0 – 4.7	6.4 – 7.0	9.5 – 9.8
C. sporogenes	5.0 – 5.8	6.0 – 7.6	8.5 – 9.0	Strepthococcus lactis	4.1 – 4.8	6.4	9.2
Erwinia carotovora	4.6	7.1	9.3	Vibrio	5.5 – 6.0		9.0
Escherichia coli	4.3 – 4.4	6.0 – 8.0	9.0 – 10	V.cholerae	-	7.5 – 8.5	-
Gluconobacter oxydans	4.0 – 4.5	5.5 – 6.0	-	V. parahaemolyticus	4.8 – 5.0	7.5 – 8.5	11.0
Lactobacillus (most)	3.0 – 4.4	5.5 – 6.0	7.2 – 8.0	Yeast	1.5 – 3.5	4.0 – 6.5	8.0 – 8.5
L. acidophilus	4.0 – 4.6	5.5 – 6.0	7.0	Hansenula	-	4.5 – 6.5	-
L. plantarum	3.5	5.5 – 6.5	8.0	Kluyveromyces	1.5 – 2.0	-	-
Leuconostoc cremoris	5.0	5.5 – 6.0	6.5	Pichia	1.5	-	-
L. oenos	-	4.2 – 4.8	-	Saccharomyces cerevisiae	2.0 – 2.4	4.0 – 5.0	-
Pediococcus cerevisiae	2.9	4.5 – 6.5	7.8	S. rouxii	1.5	3.5 – 5.5	8.5 – 10.5
Propionibacterium	4.7	6.2 – 7.0	7.5	Molds		4.5 – 6.8	8.0 – 11
Proteus vulgaris	4.4	6.0 – 7.0	8.4 - .2	Aspergillus niger	1.2	3.0 – 6.0	-
Pseudomonas (most)	5.6	6.6 – 7.0	8.0	A. oryzae	1.6 – 1.8	-	9.0 – 9.3
P. aeroginosa	5.6	6.6 – 7.0	8.0 – 9.0	Botrytis cinerea	2.5	-	7.4
Salmonella (most)	4.5 – 5.0	6.0 – 7.0	8.0 – 9.6	Mucor	-	3.0 – 6.1	9.2
				Penicillium	1.9	4.5 – 6.7	9.3
				Rhizopus nigricans	-	4.5 – 6.0	-

Source: Banwart, 1989

REDOX RANGES AND BACTERIAL ACTIVITY IN RELATION TO OXYGEN

Organism	Redox range (mV)	Activity in relation to oxygen
Pseudomonas fluorescens	+ 500 → + 100	Obligate aerobe
Staphylococcus aureus	+ 180 → -230	Facultative anaerobe
Proteus vulgaris	+ 150 → -600	Facultative anaerobe
Clostridium spp	- 30 → -550	Obligate anaerobe
Clostridium perfringens	+216 → - 230	Aerotolerant anaerobe

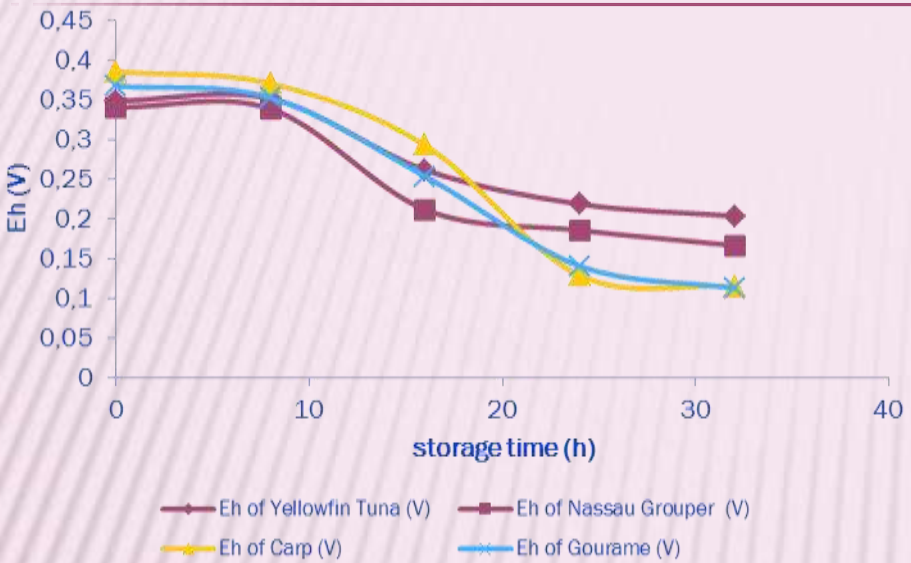
Source: Garbutt, 1997

OXYGEN REQUIREMENTS OF FOOD POISONING ORGANISM

Organism	Oxygen/ redox requirement
Salmonella sp	Facultative anaerobe
Yersinia enterocolitica	Facultative anaerobe
Escherichia coli	Facultative anaerobe
Staphyloccus aureus	Facultative anaerobe
Bacilus cereus	Facultative anaerobe
Listeria monocytogenes	Facultative anaerobe
Campylobacter	Microaerophile
Clostridium botulinum	Obligate anaerobe Oxygen is toxic
Clostridium perfringens	Obligate anaerobe Aerotolerant
Mytoxigenic moulds	Obligate aerobes

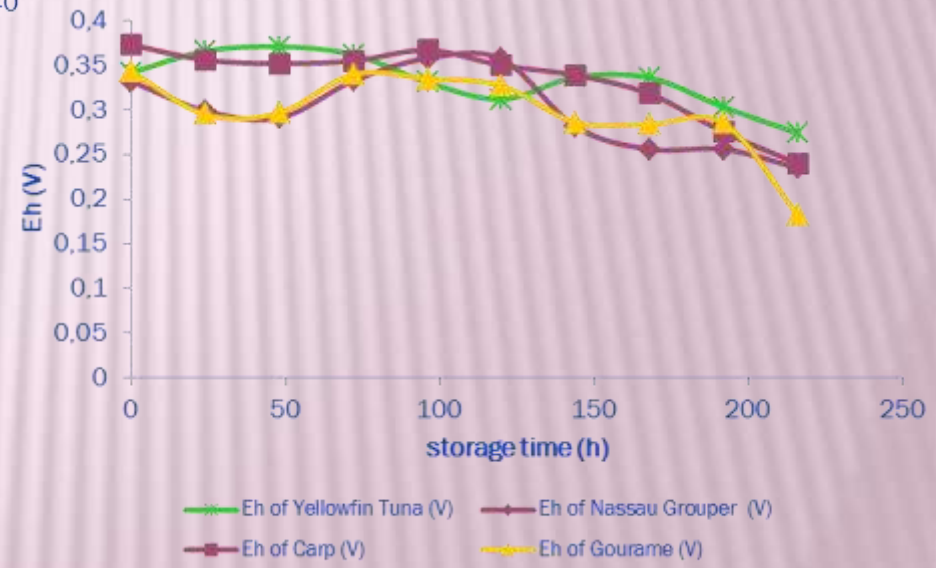
Source: Garbutt, 1997

Eh CHANGES IN TROPICAL FISH AT DIFFERENT TEMPERATURE



Ambient temperature

Source: Susanto et al., 2011

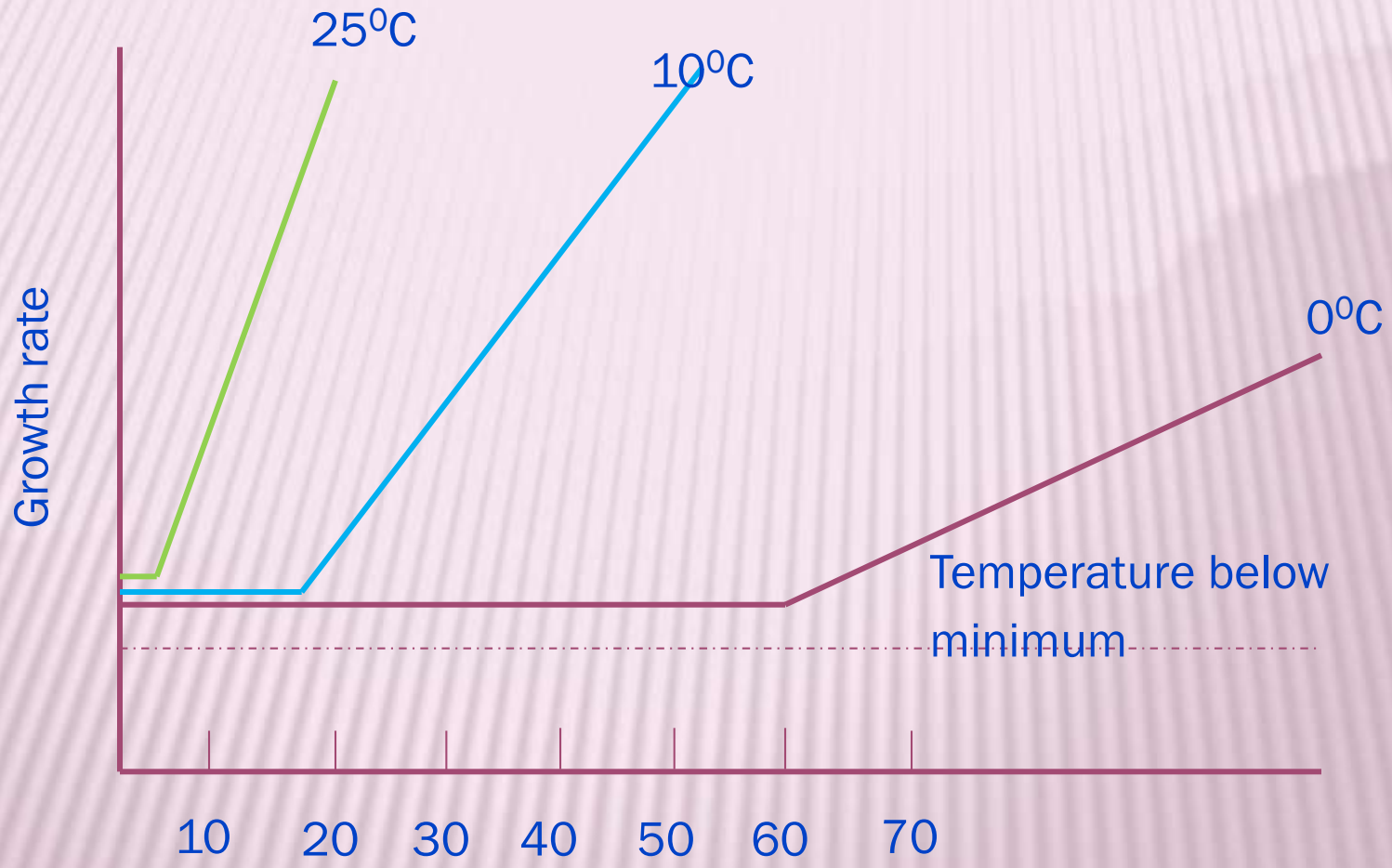


Chilled temperature

EXTRINSIC FACTORS

- Temperatures
- Gaseous atmosphere
- Rh
- Time
- Processing practices

EFFECT OF TEMPERATURE ON GROWTH RATE AN LAG PHASE



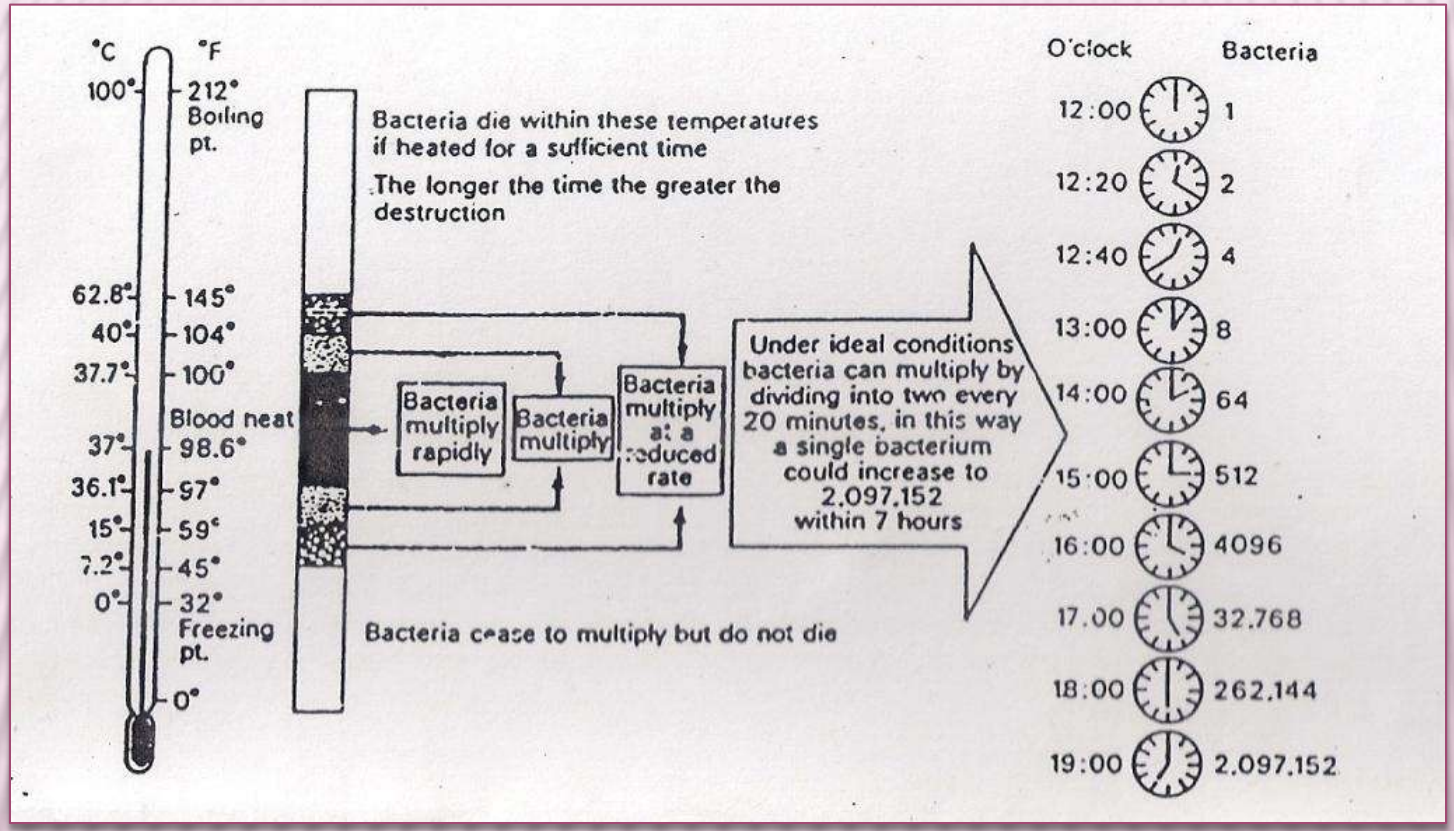
Source: Garbutt, 1997

TEMPERATURE RANGE OF SELSECTED MO

Organism	temperature			Organism	temperature		
	Min.	Opt.	Max.		Min.	Opt.	Max.
Bacteria (most)				S. aureus	5 – 10	35 – 39	44 – 48
Acetobacter	5	-	42	Streptococcus cremoris	-	25 – 30	-
Aeromonas	0 – 5	25 – 30	38 – 41	S. faecalis	5 – 10	37	49 – 51
Bacillus subtilis	10	-	47 – 50	S. lactis	10 – 15	25 – 30	0
Brevibacterium	5	-	42	S. thermophilus	20	40 – 45	52
Clostriium	0 – 45	-	60	Vibrio	-	10 – 37	-
C. botulinum	3 – 10	30 – 40	42 – 45	V. parahaemolyticus	3 – 13	35 – 37	42 – 44
C. pefringens	15 – 20	30 – 40	45 – 51	Xanthomonas	0 – 5	25 – 31	40
C. putrefaciens	0	20 – 25	30	Yersinia enterocolitica	0 – 4	-	37
C. thermosaccharolyticum	43 – 45	55 – 62	70 – 71	Molds			
Escherichia coli	3 – 10	37 – 41	48 – 50	Aspergillus niger	-	30 – 40	-
Lactobacillus (most)	5	30 – 40	53	Botrytis cinerea	-1	20	30
Leuconostoc	10	20 – 30	40	Cladosporium	-5 - -8	-	-
Micrococcus	10	25 – 30	45	Penicillium rubrum	-	25 – 28	-
Moraxella	-1 – 2	30	41 – 42	Rhizopus stolonifer	5	-	25
Propionibacterium	2 – 3	30 – 37	45	Yeast			
Proteus	10	37	43 – 45	Candida	0	-	29 – 48
Pseudomonas (most)	-7 – 4	20 – 30	31 – 43	C. lipolytica	5	25	35 – 40
P. aeroginosa	8	-	42	Hansenula	-	37 – 42	50
P. fluorescens	0 – 6	20 – 25	40	Saccharomyces	0 – 7	20 – 35	40
Salmonella (most)	5 - 10	35 – 37	46 – 49	Torulopsis	0	17 - 25	30 – 35
Staphylococcus	5 - 10	35 – 40	46 - 48				

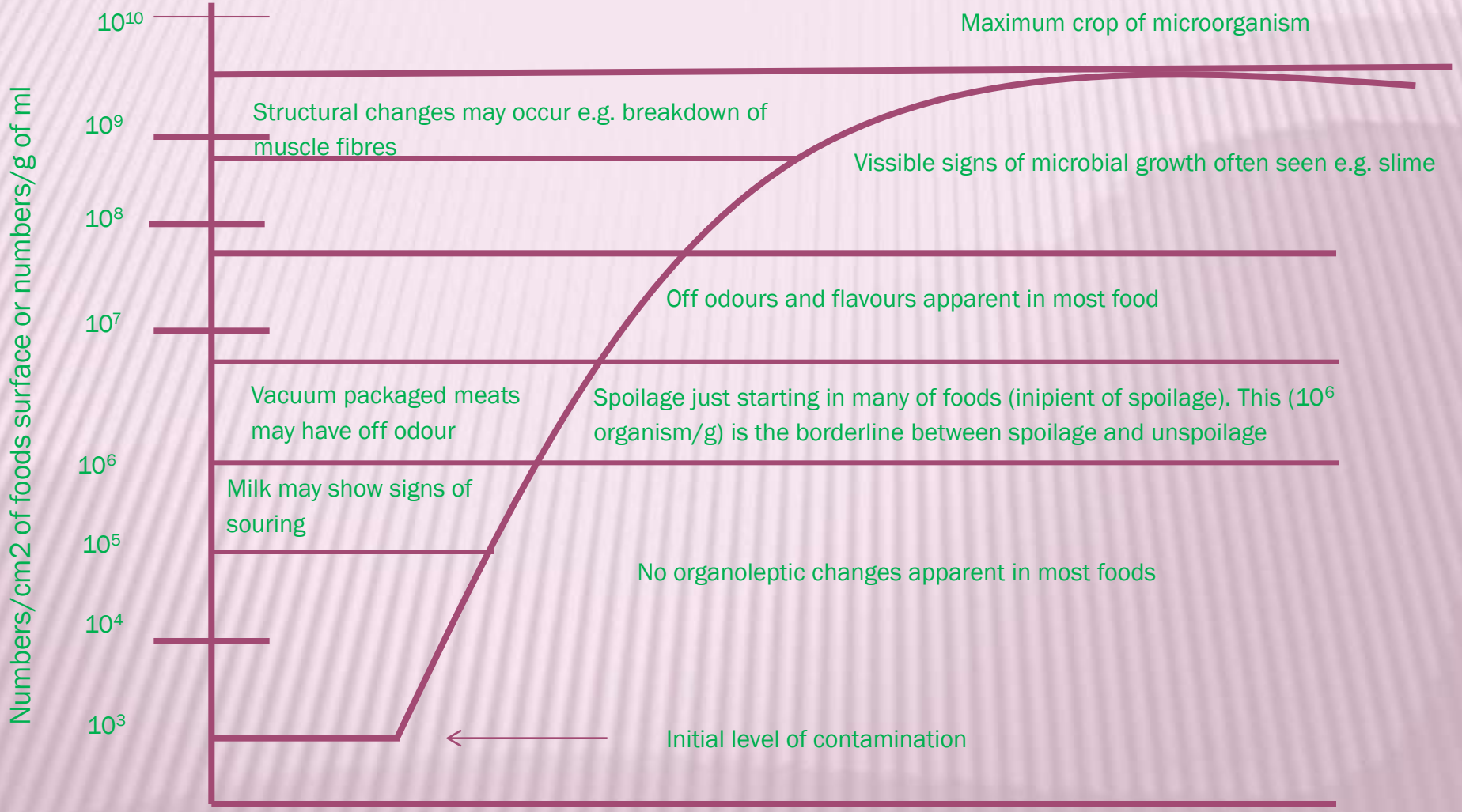
Source: Banwart, 1989

EFFECTS TEMPERATURE & TIME ON MICROBIAL GROWTH



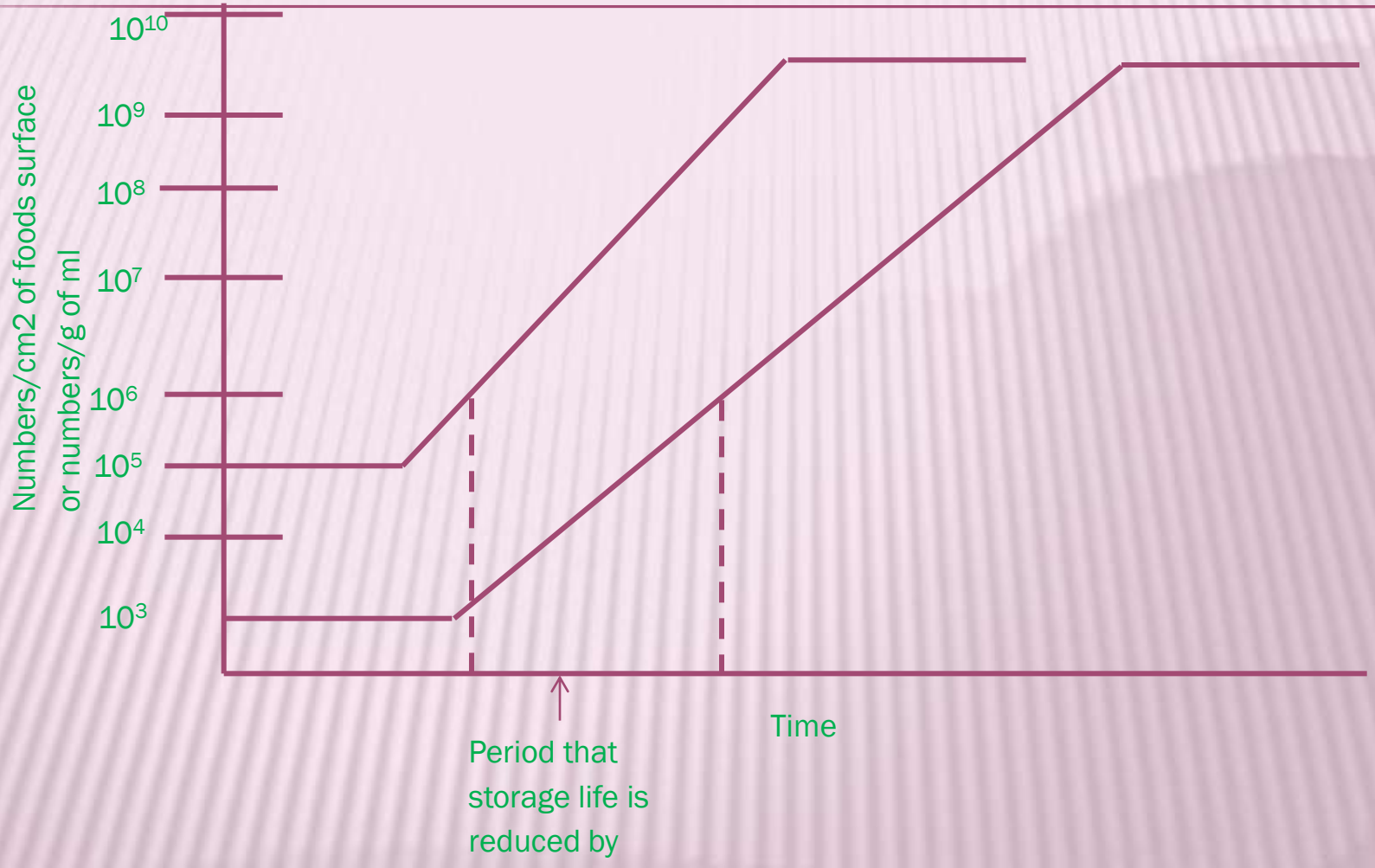
Source : Jay, 2000

GROWTH OF A BACTERIAL SPOILAGE FLORA AND ITS RELATIONSHIP TO THE SPOILAGE SYMPTOMS



Source: Garbutt, 1997

EFFECTS OF CONTAMINATION LEVEL ON STORAGE LIFE OF A FOOD

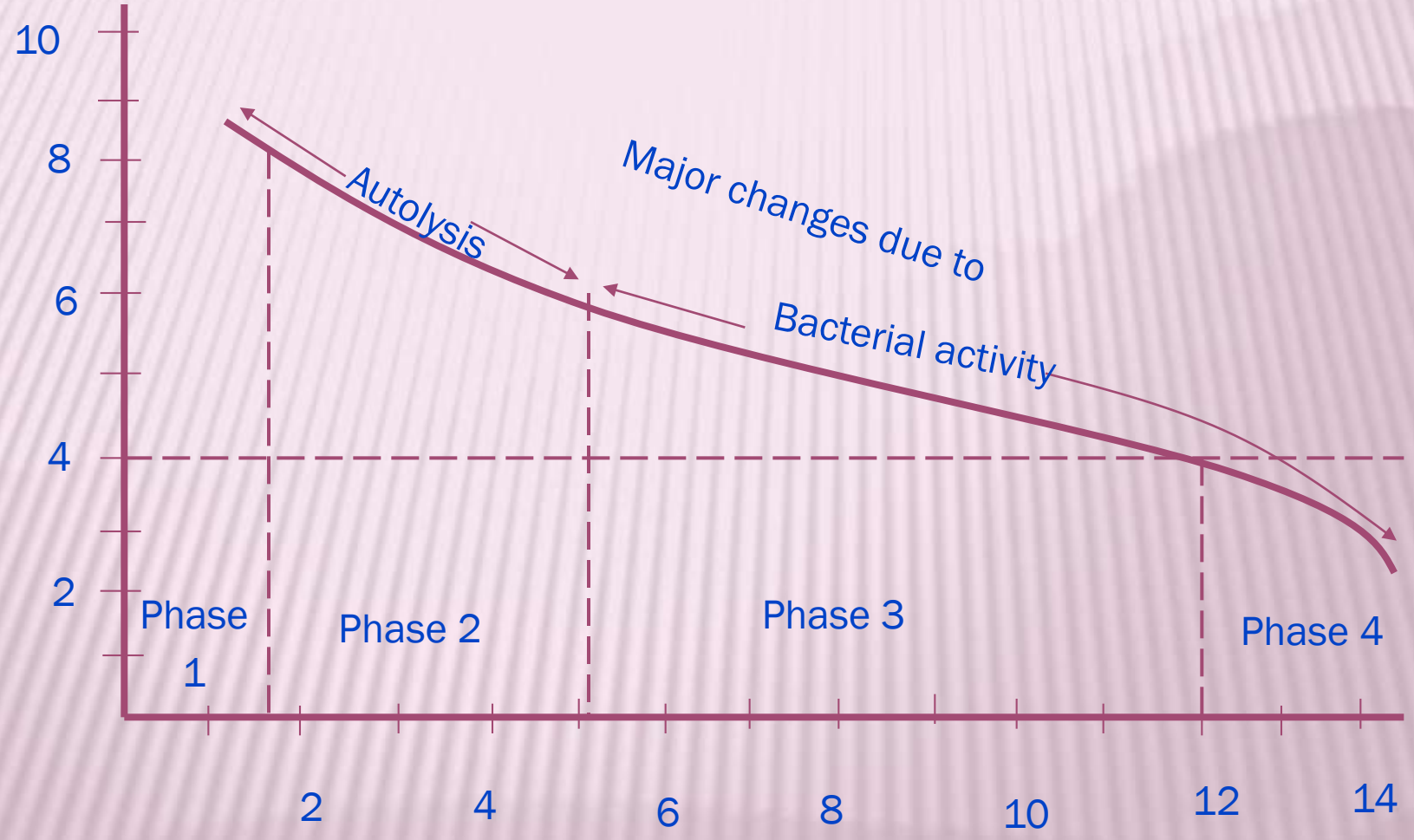


Source: Garbutt, 1997

FISH SPOILAGE

- Chemical decomposition
- Bacterial action
- oxidation

CHANGES IN SENSORY QUALITY OF ICED COD ($0^{o}C$)

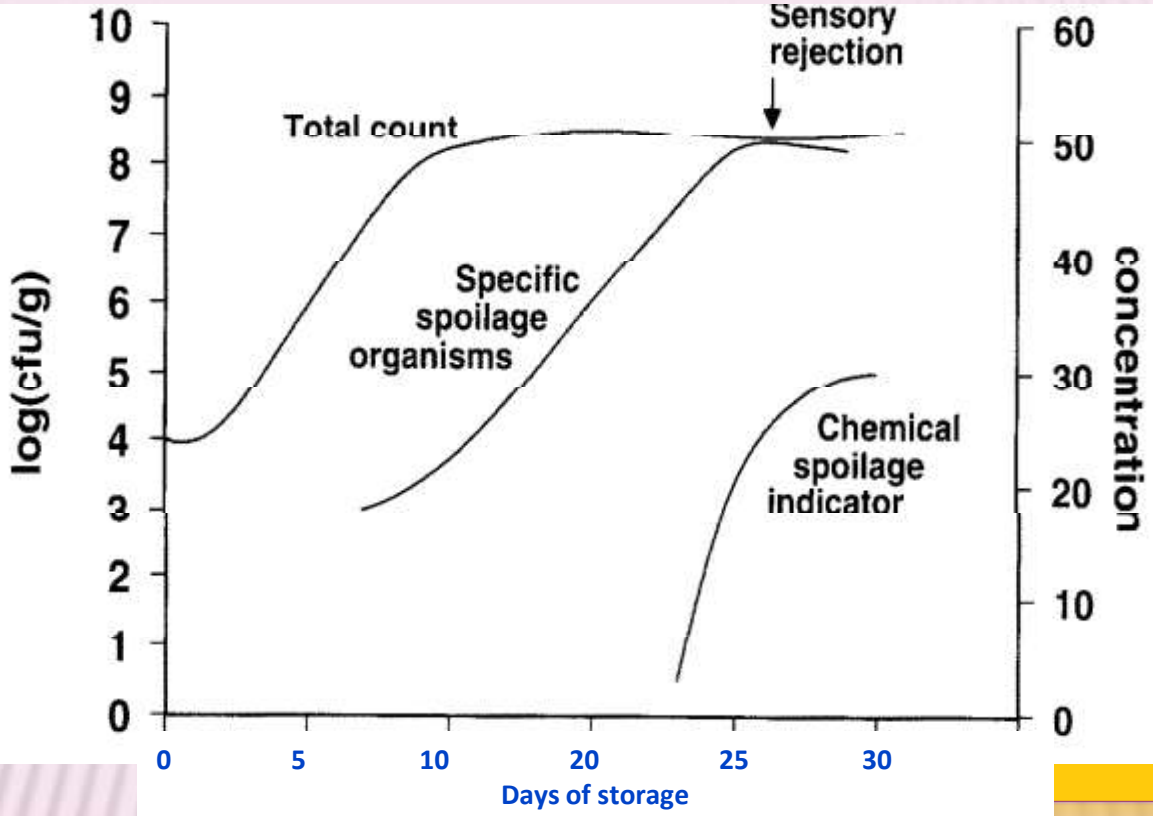


Source: Huss, 1995

THE SPOILAGE IN FISH

- under aerobic iced storage, the flora is composed almost exclusively of ***Pseudomonas*** sp. and *S. putrefaciens*. This is true for all fish and shellfish whether caught or harvested in temperate, tropical & sub tropical area.
- At ambient temperature (25”C), the microflora is dominated by mesophilic *Vibrionaceae,* the fish are caught in polluted waters, mesophilic *Enterobacteriaceae*.
- ***Shewanella putrefaciens*** is the specific spoilage bacteria of marine temperate & tropical-water fish.
- ***Pseudomonas*** sp. are the specific spoilers of iced stored tropical freshwater fish.
- motile aeromonads are the specific spoilers of aerobically stored freshwater fish

- The number of *Pseudomonas* is reduced, but S. *putreficiens* which is capable of anaerobic respiration using TMAO grows to levels of IO^6 - 10^8 cfu/g.
- *P. phosphoreum* is a marine vibrio easily isolated from intestines of various fish. This organism produces IO- 100 fold more TMA per cell than S. putrefaciens. but does not cause off-odours as foul as S. *putrefuciens.*
- The spoilage of vacuum-packed fish from temperate marine waters is caused by these two bacteria and differences in initial numbers of S. *putrefuciens* and ***P. phosphoreum.***
- CO,-packing of marine fish from temperate waters inhibits the development of the respiratory organisms like ***Pseudomonas*** and S. putrerfaciens and their numbers rarely exceed 105~10" cfu/g.
- neither ***Psrudomonas*** nor ***P. phosphoreum*** produce significant amounts of H_2S.



Model of changes in total count (TVC), specific special organism (SSO), and chemical spoilage indices during chilled of fish storage (Gram and Huss, 1996)

PROPERTIES OF FISH SPOILAGE

Test	Group			
	P. putrefaciens	Alteromonas[1]	Alteromonas[2]	Pseudomonas[3]
Fish spoilage	Strong odour	Strong odours (D)	Odours (D)	Strong odours
Shrimp spoilage	Mostly strong odour	Strong odours (D)	Strong odours (R)	Strong odours
H2S cystein	+	+ / - (2/10)	+/-	-
Gelatin liquefied	+	+	+	+
TMAO reduced	+	+	-	-
Hypoxanthine from IMP	+	+	+	-
[1] non-defined spoiler	[2] pseudomonad-like shrimp spoilers	[3] pseudomonas groups I and II	* D = Delayed	* R = rapid

Source: Van Spreekens, 1977

MICROORGANISM ASSOCIATED WITH THE SPOILAGE OF CHILLED FOODS

Bacteria	Yeasts	Moulds
Pseudomonas	Candida	Penicillium
Alteromonas	Torulopsis	Aspergillus
Shewanella	Saccharomyces	Cladosporium
Bacilus	Debariomyces	Botrytis
Clostridium	Rhodotorula	Alternaria
Lactobacillus		Trichsoporon
Brochothrix		

Source: Garbutt, 1997

CAUSES OF FISH SPOILAGE

Signs of spolage	Causes of fish spoilage			
	Microbioloical	Chemical (oxidation)	Autolytic	Physical
Off odours / off flavour	+	+	+	-
Slime formation	+	-	-	-
Gas formation	+	-	-	-
Disolouration	(+)	+	+	+
Change of texture	(+)	-	+	+

Source: Huss, 1995

PERCENTAGE ORGANISM FROM ICE FISH FILLETS PRODUCING TMA & TVB

Days in ice	TMA		TVB	
	Pseudomonas	Achromobacter	Pseudomonas	Achromobacter
0	12	14	75	43
4	8	5	32	11
8	22	15	45	45
12	21	18	42	29
118	48	0	95	100

Source: Lisson, 1979

SPECIFIC SPOILAGE BACTERIA OF FRESH AND PACKED FISH STORED CHILLED (<4"C) OR IN ICE (GRAM AND HUSS, 1996)

Atmosphere	Specific spoilage organisms of fresh, chilled fish			
	Temperate waters		Tropical waters	
	Marine	Fresh	Marine	Fresh
aerobic	S. putrefaciens	Pseudomonas spp	S. putrefaciens	Pseudomonas spp.'
	P.reudomonas spp.		Pseudomonas spp.	
vacuum	S. putrt$ciensb	Gram-positive bacteriae	Lactic acid bacteria	Lactic acid bacteria?
	P. phosphoreum	Lactic acid bacteria	/ others	
CO2	P. phosphoreum	Lactic acid bacteriad	Lactic acid bacteria / TMAO reducing bacteria	Lactic acid bacteria'?/TMAO reducing bacteria'

Source: Gram an Huss, 1996

BACTERIAL SPOILAGE COMPOUNDS

Specific spoilage bacteria	Spoilage compounds
Shewanella putrifaciens	TMA, H_2S, CH_3SH, $(CH_3)_2S$, HX
Photobacterium phosphoreum	TMA, HX
Pseudomonas spp.	Ketones, aldehydes, esters, non -H_2S sulphides
Vibrionacaea	TMA, H_2S
Aerobic spoilers	NH_3, acetic, butyric and propionic acid

Note:
TMA: Trimethylamine; H_2S: Hydrogen sulphide; CH_3SH: Methylmercarptan; $(CH_3)_2S$: Dimethylsulphide; HX = Hypoxanthine; NH_3: Ammonia

Source: Church, 1998 *in* Ghaly *et al.*, 2010

MICROBIAL FLORA OF CRUSTACEANS

- Shrimp > content of free amino acids than fish & contain chathepticklike enzymes that rapidly breakdown proteins.
- Spoiled microflora of crustaceans : Pseudomonads, Acinetobacter-Moraxella, Yeast spp.
- 0oC → *Pseudomonas*; 5.6oC → *Moraxella*; 16.7 ^{o}C & 22.2 ^{o}C → *Proteus*
- Ice storage → increase Acinetobacter/Moraxella (> 80%)
- Minor in *Shewanella* spp & *Pseudomonads* → *> NH3, TMA, Hx & acetic acids.*
-

MOST PREDOMINANT BACTERIA IN SHRIMP HELD TO SPOILAGE

Temperature (°C)	Days held	Organism
0	13	*Pseudomonas*
5.6	9	*Moraxella*
11.1	7	*Moraxella*
16.7	5	*Proteus*
22.2	3	*Proteus*

Source: Gram and Huss, 1996

MICROBIAL FLORA OF MOLLUSKAN

- Molluskan chem composition differ from both teleostei & crustaceans → (> KH & < proteins).
- Free arginine, aspartic, & glutamic acid > in fish.
- Mo in spoiled oysters : *Serratia, Pseudomonas, Proteus, Clostridium, Bacillus, Escherichia, Enterobacter, Shewanella, Lactobacillus, Flavobacterium, & Micrococcus.*
- Late stage of oyster spoilage : Pseudomonas, Acinetobacter-Moraxella spp. Predominate, with enterococci, lactobacillii & yeasts.
- Chilled temperature: dominated by Acinetobacter/Moraxella spp

BACTERIA FISH AND SHELLFISH (%)

Fish type	Pseudomonas	Vibrio	Achromobacter	Coryneform	Others
North sea fish	5	-	56	-	39
Haddock	26	2	45	4	23
Flatfish	20	13	30	17	20
Pescada	32	-	35	4	29
Shrimp (north pacific)	10	-	47	3	40
Shrimp (Gulf ocean)	22	2	15	40	21
Shrimp (pond culture)	-	5	2	83	13
Scampi (UK)	3	-	11	81	5
Mullet (Queensland)	18	-	9	12	19

Source: Lisson, 1979

MICROBIAL SPOILAGE OF FISH

Microbiological activity	Sensory manifestation
Breakdown of food components	Production of off flavour and oddour
Production of extracellular polysacharide material	Slime formation
Growth of per se of moulds, bacteria, yeast	Large visble of pigmented or non pigmented colonies
CO2 from carbohydrate or aminoacids	Production of gas
Production of diffusible pigment	Discolouration

Source: Gram and Huss, 1996

GENERA BACTERIA, YEASTS, & MOLDS MOST OFTEN FOUND ON FRESH & SPOILED FISH & OTHER SEAFOODS

Bacteria	Gram	Prevalence	Yeasts	Prevalence	Molds	Prevalence
Acinetobacter	-	X	Candida	XX	Aspergillus	X
Aeromonas	-	XX	Cryptococcus	XX	Aureobasidium (Pullularia)	XX
Alcaligenes	-	X	Hansenula	X	Penicillium	X
Bacillus	+	X	Pichia	X	Scopulariopsis	X
Corynebacterium	+	X	Rhodotorula	XX		
Enterobacter	-	X	Sporobolomyces	X		
Enterococcus	+	X	Trichosporon	X		
Escherichia	-	X				
Flavobacterium	-	X				
Lactobacillus	+	X				
Listeria	+	X				
Microbacterium	+	X				
Moraxella	-	X				
Photobacterium	-	X				
Pseudomonas	-	XX				
Psychrobacter	-	X				
Shewanella	-	XX				
Vibrio	+	XX				
Weissella		X				
Note: X = known to occur; XX = most requently reported						

Source: Jay, 2000

MICROBIAL CHANGES IN PACIFIC HAKE STORED AT 5^{0}C

Microrganism	Microbial population after incubation (%)			
	0 day	5 days	8 days	14 days
Pseudomonas				
Type 1	14.0	7.3	2.7	15.1
Type II	14.0	52.4	53.4	77.4
Type III or IV	3.5	12.2	31.5	7.5
Acineobacter-Moraxela				
Acinetobacter	31.6	17.0	8.2	0
Moraxella	19.3	9.8	2.7	0
Flavobacterium	17.6	0	0	0
Coliforms	0	1.2	1.4	0
Microbial count of sample	1.5×10^4	3.4×10^2	9.3×10^6	2.7×10^6
Number of microorganisms identified	57	82	73	53

Source: Gram and Huss, 1996

SOME MICROBIAL DEFECTS OF SEAFOODS

Product	Defect	Microorganism
Fish		
Fresh	Off-odor	*Pseudomonas, Alteromonas, Acinetobacter, Vibrio, Aeromonas, Moraxella, Proteus,*
	Fruity	*Psedomonas*
	Ammoniacal	*Psedomonas, Alteromonas*
	H_2S odor	*Pseudomonas, Alteromonas*
Salted	Pink	*Halobacterium, Halococcus*
	Dun (red growth)	*Hemispora stellata, Sporendonema epizoum*
	Cheesy, putrefactive	Red halophilic bacteria
Crayfish	Sweet to foul odor	*Pseudomonas, Lactobacillus, Coryneforms*
Oysters	Pink	Yeasts (*Rhodotorula*)
Hrimp	Off-odor	*Pseudomonas*
Squid	Yellow discoloration	*P. putida*
	Red discoloration	*Serratia marcescens*

Source: Banwart, 1989

SPOILAGE OF FISHERIES PRODUCTS

- Lightly preserved fish products
- Salt curing & fermentation
- Heat treated products
- Smoking
- Packaged fish

LIGHTLY PRESERVED FISH PRODUCTS (LPFP)

- Lactic acid bacteria dominate in LPFP
- *Lactobacillus curvatus was* the most common species occurring, but also *Lb. sake/ bavarius, Lb. Plantarum, Carnobacterium*, and *Leuconostoc* sp. were present in smaller numbers.
- Spoiled LPFP may contain (10^5cfu/g) or high (10^6- 10^7 cfu/g) levels of *Enterobucteriuceae, Brochotrix thermosphnctu,* yeasts and *Photobacterium phosphoreum.*
- The *Enterobucteriuceue* growing in lightly preserved fish products have been identified as psychrotrophic *Hafnia alvei, Serratia liquefaciens* or *Enterobacter* sp. -→ produce H2S, off odour & reduce TMAO.

SALTED FISH

- Predominantly gram negative flora of fish sensitive to high salt concentration.
- High concentration salt ➔ *Halobacterium & Halococcus* (20 % NaCl)
- *Sporendonema expizoum & Oospora* ➔ *dun* spoilage
-

SMOKED FISH

- In hot smoking → micrococci & Bacillus sp
- Brined cold smoke → Pseudomonas; slight in salt → predominating micrococci
- Penicillium & Aspergillus spp → causing spoilage in smoked fish.
- Cold smoked salmon was dominated by lactic acid bacteria (10^7-10^9 cfu/g); (b) dominated by lactic acid bacteria and *Enterobacteriaceae* (10^7-10^8 cfu/g); and (c) dominated by *Photobacterium phosphoreum* (10^6 - 10^7 cfu/g) with occasional high levels of lactic acid bacteria.

FUNGI ON SALTED N SMOKED FISH

Genus	Source	Author
Aspergills sp	Smoked fish	Graikoski (1973)
Oospora Nikitinski	Salted fish	Malevich (1936)
Oospora spp	Dun of salted fish	Frank and Hess (1941)
Penicillium sp	Smoked fish	Graikoski (1973)
Sporendonema epizoum	Salted fish	Van Klavern and Lequendre (1965)
Sporendonema spp	Dun of salted fish	Frank and Hess (1941)

Source: Lisson, 1979

PACKAGED FISH

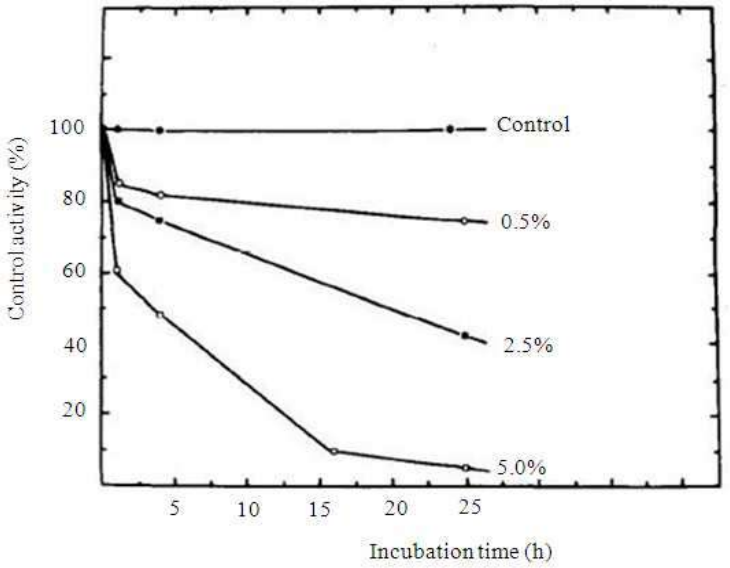
- Vacuum-packaging -→ botulism
- Protection againts C. botulinum type E → > 4.5 % NaCl & 50 µg/kg sodium nitrite
- Herring stored n CO2 storage → Lactobacillus spp become predominant.

HEAT TREATED FISH PRODUCTS

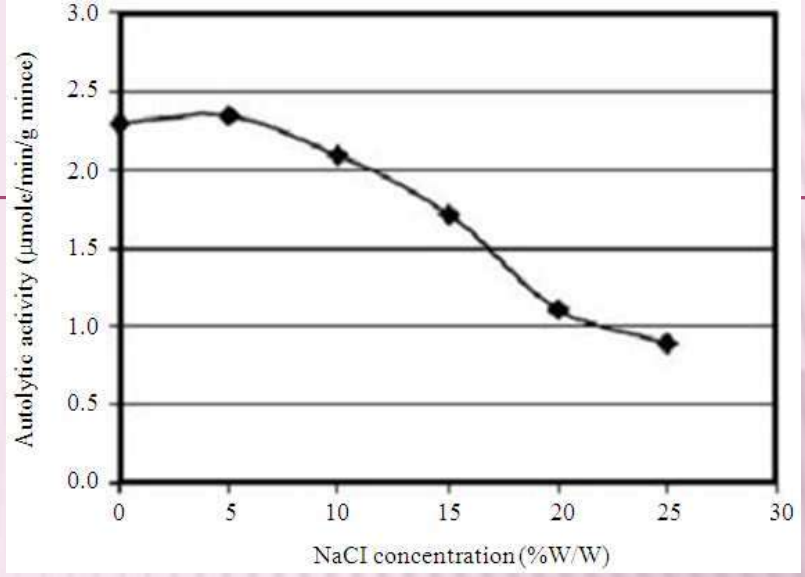
- Clostridum botulinum & gram positve bacteria were found in pasteurized crab meat.

FISH SPOILAGE PRESERVATION TECHNIQUE

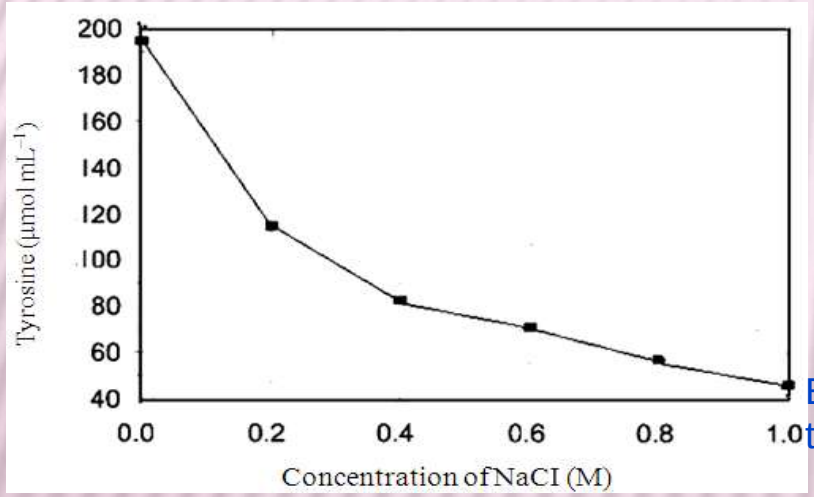
- Low temperature storage
- Controlling Aw
- Controlling fish spoilage
- Gutting
- Addition of sodium chloride
- Addition of acids
- Phenolic antioxidants
- Controlling microbial spoilage
- Nitrites, shulphites, lactic acids & benzoics acids
- CO_2 packaging on fish products



The effect of NaCl on cathepsin (Reddi *et al*., 1972)



Effect of NaCl on autolytic activity in Indian anchovy (Siringan *et al*., 2006)



Effect of NaCl concentration on autolytic activity of tilapia surimi at 65°C (Yongsawatdigul *et al*., 2000)

THANK YOU FOR ATTENTION